Form PTO-1449

INFORMATION DISCLOSURE CITATION IN AN APPLICATION

(Use several sheets if necessary)

ATTY DOCKET NO.	APPLICATION NO.
0020-4764P	NEW
APPLICANT	
Hiroki NAKAJIMA et al	
FILING DATE	**GROUP**
October 27, 2000	1638

U.S. PATENT DOCUMENTS

EXAMINER INITIAL	DOCUMENT NUMBER	DATE	NAME	CLASS	SUB CLASS	FILING DATE IF APPROPRIATE

FOREIGN PATENT DOCUMENTS

	DOCUMENT NUMBER	DATE	COUNTRY	CLASS	SUB CLASS	TRANSLATION YES	TRANSLATION NO
DK	WO 95/46059	1995-12-21	WO				
	WO 97/32011	1997-09-04	WO				
	EP 0770682A2	1997-05-02	EP				
	WO 97/04088	1997-02-06	WO				
	WO 98/33927	1998-08-06	WO				
DK	WO 98/43027	1998-01-05	~~GERMANY~~ WO			ABS	
	WO 98/42852	1998-10-01	WO			Abs	

OTHER DOCUMENTS (Including Author, Title, Date, Pertinent Pages, etc.)

DK	Kruse, E. et al; PLANTA (HEIDELBERG) 1995, vol. 196, no. 4, 1995, pages 796-803, ~~XP 000916336~~
	Kruse, E. et al; PLANT MOLECULAR BIOLOGY DEC., 1997, vol. 35, no. 6, pages 1053-1056, ~~XP002076483~~.
DK	Madsen, O. et al; PLANT MOLECULAR BIOLOGY 1993, vol. 23, no. 1, 1993, pages 35-43, ~~XP000916344~~.

EXAMINER	DATE CONSIDERED
David Kruse	9 January 2007

EXAMINER: Initial if citation considered, whether or not citation is in conformance with M.P.E.P. 609; Draw line through citation if not in conformance and not considered. Include copy of this form with next communication to applicant.

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International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : C12N 9/02, 15/82 — A1

(11) International Publication Number: **WO 97/32011**

(43) International Publication Date: 4 September 1997 (04.09.97)

(21) International Application Number: PCT/US97/03313

(22) International Filing Date: 27 February 1997 (27.02.97)

(30) Priority Data:

60/012,705	28 February 1996 (28.02.96)	US
60/013,612	28 February 1996 (28.02.96)	US
60/020,003	21 June 1996 (21.06.96)	US

(81) Designated States: AU, BA, BB, BG, BR, BY, CA, CN, CU, CZ, FI, GE, GH, HU, JP, KG, KR, KZ, LC, LK, LV, MD, MG, MN, MW, NO, NZ, PL, RO, RU, SD, SK, TJ, UA, US, UZ, VN, YU, European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

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(54) Title: DNA MOLECULES ENCODING PLANT PROTOPORPHYRINOGEN OXIDASE AND INHIBITOR-RESISTANT MUTANTS THEREOF

(57) Abstract

The present invention provides novel DNA sequences coding for plant protoporphyrinogen oxidase (protox) enzymes from soybean, wheat, cotton, sugar beet, grape, rice and sorghum. In addition, the present invention teaches modified forms of protox enzymes that are herbicide tolerant. Plants expressing herbicide tolerant protox enzymes taught herein are also provided. These plants may be engineered for resistance to protox inhibitors via mutation of the native protox gene to a resistant form or they may be transformed with a gene encoding an inhibitor-resistant form of a plant protox enzyme.

Soybean Protox-1, in the pBluescript SK vector, was deposited December 15, 1995 as pWDC-12 (NRRL #B-21516).

An alignment of the predicted amino acid sequences of the respective proteins encoded by the sequences shown in SEQ ID NOS: 2, 6, 10, 12, 15, 17, 19, 21, 23 and are set forth in Table 1. An alignment of the predicted amino acid sequences of the respective proteins encoded by the sequences shown in SEQ ID NOS: 4 and 8 are set forth in Table 2.

TABLE 1

Comparison of Protox-1 Amino Acid Sequences from Arabidopsis ("Arabpt-1"; SEQ ID NO:2), Maize ("Mzpt-1"; SEQ ID NO:6), Wheat ("Wtpt-1"; SEQ ID NO:10), Soybean ("Soybeanpt-1"; SEQ ID NO:12), Cotton ("Cottonpt-1"; SEQ ID NO:16), Sugar beet ("Sugpt-1"; SEQ ID NO:18), Rape ("Rapept-1"; SEQ ID NO:20), Rice ("Ricept-1"; SEQ ID NO:22), and Sorghum ("Sorghumpt-1"; SEQ ID NO:24)

Alignment is performed using the PileUp program (GCG package, University of Wisconsin, Madison, WI). Positions that may be modified according to the teachings herein to confer or enhance inhibitor resistance are shown in bold type.

```
              1                                                   50
   Rapept-1   .......... .......... MDLSLLRP.. QPFLSPFSNP FPRSRPYKPL
   Arabpt-1   .......... .......... MELSLLRPTT QSLLPSFSKP NLRLNVYKPL
Sorghumpt-1   .......... .......... .......... .......... ..........
     Mzpt-1   .......... .......... .......... .......... ..........
     Wtpt-1   .......... .......... .........M ATATVAAASP LRGRVTGRPH
   Ricept-1   .......... .......... .......... .......... ..........
 Cottonpt-1   .......... ......MTAL IDLSLLRSSP SVSPFSIPHH QHPPRFRKPF
 Soybeanpt1   ........MV SVFNEILFPP NQTLLRPSLH SPTSFFTSPT RKFPRSRPNP
    Sugpt-1   MKSMALSNCI PQTQCMPLRS SGHYRGNCIM LSIPCSLIGR RGYYSHKKRR

              51                                   GXGXXG         100
   Rapept-1   NLRCSVSGGS VVGSSTIEGG GGGKTVTADC VIVGGGISGL CIAQALVTKH
   Arabpt-1   RLRCSVAGGP TVGSSKIEGG GGT.TITTDC VIVGGGISGL CIAQALATKH
Sorghumpt-1   .......... .......... .......... .......... ..........
     Mzpt-1   .......... .......... .......ADC VVVGGGISGL CTAQALATRH
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Wtpt-1      RVRPRCATAS SATETPAAPG VRL...SAEC VIVGAGISGL CIAQALATRY
Ricept-1    .......... .......... .......... .......... ..........
Cottonpt-1  KLRCSLAEGP TISSSKIDGG ESS...IADC VIVGGGISGL CIAQALATKH
Soybeanpt1  ILRCSIAEES TASPPKTR.. DSA...PVDC VVVGGGVSGL CIAQALATKH
Sugpt-1     MSMSCSTSSG SKSAVKEAGS GSGAGGLLDC VIVGGGISGL CIAQALCTKH
                                              GxGxxG

            101                                                  150
Rapept-1    PDA..AKNVM VTEAKDRVGG NIIT..REEQ GFLWEEGPNS FQPSDPMLTM
Arabpt-1    PDA..APNLI VTEAKDRVGG NIIT..REEN GFLWEEGPNS FQPSDPMLTM
Sorghumpt-1 .......... .......... ..STVERPEE GYLWEEGPNS FQPSDPVLSM
Mzpt-1      ..G..VGDVL VTEARARPGG NITTVERPEE GYLWEEGPNS FQPSDPVLTM
Wtpt-1      ..G..VSDLL VTEARDRPGG NITTVERPDE GYLWEEGPNS FQPSDPVLTM
Ricept-1    .......... .......... .......... .......... ..........
Cottonpt-1  RDV..ASNVI VTEARDRVGG NITTVER..D GYLWEEGPNS FQPSDPILTM
Soybeanpt1  ..A..NANVV VTEARDRVGG NITTMER..D GYLWEEGPNS FQPSDPMLTM
Sugpt-1     SSSSLSPNFI VTEAKDRVGG NIVTVE..AD GYIWEEGPNS FQPSDAVLTM

            151                                                  200
Rapept-1    VVDSGLKDDL VLGDPTAPRF VLWNGKLRPV PSKLTDLPFF DLMSIGGKIR
Arabpt-1    VVDSGLKDDL VLGDPTAPRF VLWNGKLRPV PSKLTDLPFF DLMSIGGKIR
Sorghumpt-1 AVDSGLKDDL VFGDPNAPRF VLWEGKLRPV PSKPADLPFF DLMSIPGKLR
Mzpt-1      AVDSGLKDDL VFGDPNAPRF VLWEGKLRPV PSKPADLPFF DLMSIPGKLR
Wtpt-1      AVDSGLKDDL VFGDPNAPRF VLWEGKLRPV PSKPGDLPFF SLMSIPGKLR
Ricept-1    .......... .......... .......... .......... ..........
Cottonpt-1  AVDSGLKDDL VLGDPNAPRF VLWEGKLRPV PSKPTDLPFF DLMSIAGKLR
Soybeanpt1  VVDSGLKDEL VLGDPDAPRF VLWNRKLRPV PGKLTDLPFF DLMSIGGKIR
Sugpt-1     AVDSGLKDEL VLGDPNAPRF VLWNDKLRPV PSSLTDLPFF DLMTIPGKIR

            201                                                  250
Rapept-1    AGFGAIGIRP SPPGREESVE EFVRRNLGDE VFERLIEPFC SGVYAGDPAK
Arabpt-1    AGFGALGIRP SPPGREESVE EFVRRNLGDE VFERLIEPFC SGVYAGDPSK
Sorghumpt-1 AGLGALGIRP PAPGREESVE EFVRRNLGAE VFERLIEPFC SGVYAGDPSK
Mzpt-1      AGLGALGIRP PPPGREESVE EFVRRNLGAE VFERLIEPFC SGVYAGDPSK
Wtpt-1      AGLGALGIRP PPPGREESVE EFVRRNLGAE VFERLIEPFC SGVYAGDPSK
Ricept-1    .......... .......... .......... .......... ..........
Cottonpt-1  AGFGAIGIRP PPPGYEESVE EFVRRNLGAE VFERFIEPFC SGVYAGDPSK
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Soybeanpt1  AGFGALGIRP PPPGHEESVE EFVRRNLGDE VFERLIEPFC SGVYMGDPSK
   Sugpt-1  AALGALGFRP SPPPHEESVE HFVRRNLGDE VFERLIEPFC SGVYMGDPAK

                251                                              300
   Rapept-1 LSMKAAFGKV WKLEENGGSI IGGAFKAIQA KNKAPKTTRD PRLPKPKGQT
   Arabpt-1 LSMKAAFGKV WKLEQNGGSI IGGTFKAIQE RKNAPKAERD PRLPKPQGQT
Sorghumpt-1 LSMKAAFGKV WRLEEAGGSI IGGTIKTIQE RGKNPKPPRD PRLPKPKGQT
   Mzpt-1   LSMKAAFGKV WRLEETGGSI IGGTIKTIQE RSKNPKPPRD ARLPKPKGQT
   Wtpt-1   LSMKAAFGKV WRLEEIGGSI IGGTIKAIQD KGKNPKPPRD PRLPAPKGQT
   Ricept-1 RALKAAFGKV WRLEDIGGSI IGGTIKTIQE RGKNPKPPRD PRLPTPKGQT
Cottonpt-1  LSMKAAFGRV WKLEEIGGSI IGGTFKTIQE RNKTPKPPRD PRLPKPKGQT
Soybeanpt1  LSMKAAFGKV WKLEKNGGSI IGGTFKAIQE RNGASKPPRD PRLPKPKGQT
   Sugpt-1  LSMKAAFGKV WKLEQKGGSI IGGTLKAIQE RGSNPKPPRD QRLPKPKGQT

                301                                              350
   Rapept-1 VGSFRKGLIM LPEAISARLG DKVKVSWKLS SITKLASGEY SLTYETPEGI
   Arabpt-1 VGSFRKGLRM LPEAISARLG SKVKLSWKLS GITKLESGGY NLTYETPDGL
Sorghumpt-1 VASFRKGLAM LPNAITSSLG SKVKLSWKLT SMTKSDGKGY VLEYETPEGV
   Mzpt-1   VASFRKGLAM LPNAITSSLG SKVKLSWKLT SITKSDDKGY VLEYETPEGV
   Wtpt-1   VASFRKGLAM LPNAIASRLG SKVKLSWKLT SITKADNQGY VLGYETPEGL
   Ricept-1 VASFRKGLIM LPDAITSRLG SKVKLSWKLT SITKSDNKGY ALVYETPEGV
Cottonpt-1  VGSFRKGLIM LPEAIANSLG SNVKLSWKLS SITKLGNGGY NLTFETPEGM
Soybeanpt1  VGSFRKGLIM LPDAISARLG NKVKLSWKLS SISKLDSGEY SLTYETPEGV
   Sugpt-1  VGSFRKGLVM LPTAISARLG SRVKLSWTLS SIVKSLNGEY SLTYDTPDGL

                351                                              400
   Rapept-1 VIVQSKSVVM TVPSHVASSL LRPLSDSAAE ALSKLYYPPV AAVSISYAKE
   Arabpt-1 VSVQSKSVVM TVPSHVASGL LRPLSESAAN ALSKLYYPPV AAVSISYPKE
Sorghumpt-1 VLVQAKSVIM TIPSYVASDI LRPLSGDAAD VLSRFYYPPV AAVTVSYPKE
   Mzpt-1   VSVQAKSVIM TIPSYVASNI LRPLSSDAAD ALSRFYYPPV AAVTVSYPKE
   Wtpt-1   VSVQAKSVIM TIPSYVASDI LRPLSIDAAD ALSKFYYPPV AAVTVSYPKE
   Ricept-1 VSVQAKTVVM TIPSYVASDI LRPLSSDAAD ALSIFYYPPV AAVTVSYPKE
Cottonpt-1  VSLQSRSVVM TIPSHVASNL LHPLSAAAAD ALSQFYYPPV ASVTVSYPKE
Soybeanpt1  VSLQCKTVVL TIPSYVASTL LRPLSAAAAD ALSKFYYPPV AAVSISYPKE
   Sugpt-1  VSVRTKSVVM TVPSYVASRL LRPLSDSAAD SLSKFYYPPV AAVSLSYPKE
```

```
                401                                                 450
      Rapept-1  AIRSECLIDG ELKGFGQLHP RTQKVETLGT IYSSSLFPNR APPGRVLLLN
      Arabpt-1  AIRTECLIDG ELKGFGQLHP RTQGVETLGT IYSSSLFPNR APPGRILLLN
   Sorghumpt-1  AIRKECLIDG ELQGFGQLHP RSQGVETLGT IYSSSLFPNR APAGRVLLLN
      Mzpt-1    AIRKECLIDG ELQGFGQLHP RSQGVETLGT IYSSSLFPNR APDGRVLLLN
      Wtpt-1    AIRKECLIDG ELQGFGQLHP RSQGVETLGT IYSSSLFPNR APAGRVLLLN
      Ricept-1  AIRKECLIDG ELQGFGQLHP RSQGVETLGT IYSSSLFPNR APAGRVLLLN
   Cottonpt-1   AIRKECLIDG ELKGFGQLHP RSQGIETLGT IYSSSLFPNR APSGRVLLLN
   Soybeanpt1   AIRSECLIDG ELKGFGQLHP RSQGVETLGT IYSSSLFPNR APPGRVLLLN
      Sugpt-1   AIRSECLING ELQGFGQLHP RSQGVETLGT IYSSSLFPGR APPGRILILS

                451                                                 500
      Rapept-1  YIGGATNIGI LSKSEGELVE AVDRDLRKML IKPSSTDPLV LGVKLWPQAI
      Arabpt-1  YIGGSTNIGI LSKSEGELVE AVDRDLRKML IKPNSTDPLK LGVRVWPQAI
   Sorghumpt-1  YIGGATNIGI VSKTESELVE AVDRDLRKML INPTAVDPLV LGVRVWPQAI
      Mzpt-1    YIGGATNIGI VSKTESELVE AVDRDLRKML INSTAVDPLV LGVRVWPQAI
      Wtpt-1    YIGGSTNIGI VSKTESDLVG AVDRDLRKML INPRAADPLA LGVRVWPQAI
      Ricept-1  YIGGSTNIGI VSKTESELVE AVDRDLRKML INPRAVDPLV LGVRVWPQAI
   Cottonpt-1   YIGGATNIGI LSKTEGELVE AVDRDLRKML INPNAKDPLV LGVRVWPKAI
   Soybeanpt1   YIGGATNIGI LSKTDSELVE TVDRDLRKIL INPNAQDPFV VGVRLWPQAI
      Sugpt-1   YIGGAKNPGI LNKSKDELAK TVDKDLRRML INPDAKLPRV LGVRVWPQAI

                501                                                 550
      Rapept-1  PQFLIGHIDL VDAAKASLSS SGHEGLFLGG NYVAGVALGR CVEGAYETAT
      Arabpt-1  PQFLVGHFDI LDTAKSSLTS SGYEGLFLGG NYVAGVALGR CVEGAYETAI
   Sorghumpt-1  PQFLVGHLDL LEAAKSALDQ GGYNGLFLGG NYVAGVALGR CIEGAYESAA
      Mzpt-1    PQFLVGHLDL LEAAKAALDR GGYDGLFLGG NYVAGVALGR CVEGAYESAS
      Wtpt-1    PQFLIGHLDR LAAAKSALGQ GGYDGLFLGG KYVAGVALGR CIEGAYESAS
      Ricept-1  PQFLIGHLDH LEAAKSALGK GGYDGLFLGG NYVAGVALGR CVEGAYESAS
   Cottonpt-1   PQFLVGHLDL LDSAKMALRD SGFHGLFLGG NYVSGVALGR CVEGAYEVAA
   Soybeanpt1   PQFLVGHLDL LDVAKASIRN TGFEGLFLGG NYVSGVALGR CVEGAYEVAA
      Sugpt-1   PQFSIGHFDL LDAAKAALTD TGVKGLFLGG NYVSGVALGR CIEGAYESAA

                551       563
      Rapept-1  QVNDFMSRYA YK*
      Arabpt-1  EVNNFMSRYA YK*
```

```
Sorghumpt-1  QIYDFLTKYA YK*
    Mzpt-1   QISDFLTKYA YK*
    Wtpt-1   QVSDFLTKYA YK*
   Ricept-1  QISDYLTKYA YK*
 Cottonpt-1  EVKEFLSQYA YK*
 Soybeanpt1  EVNDFLTNRV YK*
   Sugpt-1   EVVDFLSQYS DK*
```

TABLE 2

Comparison of the Arabidopsis (SEQ ID NO:4) and
Maize (SEQ ID NO:8) Protox-2 Amino Acid Sequences

Identical residues are denoted by the vertical bar between the two sequences. Alignment is performed using the GAP program described in Deveraux *et al., Nucleic Acids Res. 12*:387-395 (1984).

Percent Similarity: 75.889 Percent Identity: 57.905
Protox-2.Pep x Mzprotox-2.Pep

```
  1 ..........................MASGAVAD.HQIEAVSGKRVAV 21
                               .|  |:|: .:  |..:.|||
  1 MLALTASASSASSHPYRHASAHTRRPRLRAVLAMAGSDDPRAAPARSVAV 50

 22 VGAGVSGLAAAYKLKSRGLNVTVFEADGRVGGKLRSVMQNGLIWDEGANT 71
    ||||||||||||:|: .|:|||||||.:|.|||:|.  :.|::|||||||
 51 VGAGVSGLAAAYRLRQSGVNVTVFEAADRAGGKIRTNSEGGFVWDEGANT 100

 72 MTEAEPEVGSLLDDLGLREKQQFPISQKKRYIVRNGVPVMLPTNPIELVT 121
    |||:| |.:.|:|||||.:|||:| ||.|||||::|.|.::|.:||.|:.
101 MTEGEWEASRLIDDLGLQDKQQYPNSQHKRYIVKDGAPALIPSDPISLMK 150

122 SSVLSTQSKFQILLEPFLWKK....KSSKVSDASAEESVSEFFQRHFGQE 167
    ||||||.||:.:::||||:||    .|:|||:.  .|||:.| :|||||.|
151 SSVLSTKSKIALFFEPFLYKKANTRNSGKVSEEHLSESVGSFCERHFGRE 200
```